

**Amendments to the Claims:**

Please amend Claims 1 and 2. The Claim Listing below will replace all prior versions of the claims in the application:

**Claim Listing:**

1. (Currently Amended) An isolated complex comprising heparin and <sup>the</sup> the heparin binding proteins, platelet factor 4 and thrombospondin-1 wherein each of the heparin binding proteins is an intact protein isolated from human platelets or produced using recombinant means, or a biologically active fragment prepared from a protein isolated from human platelets, or a recombinant protein, a variant recombinant protein, a synthetic peptide, or a chimeric protein.
2. (Currently Amended) The complex according to Claim 1 wherein the heparin binding proteins are isolated from ~~mammalian~~ human blood.
3. (Original) The complex according to Claim 1 wherein the platelet factor 4 is an isolated protein selected from the group consisting of: a protein isolated from human platelets, a fragment prepared from a protein isolated from human platelets, a recombinant human protein, a synthetic peptide, a recombinant human variant protein and a chimeric human protein.
4. (Original) The complex according to Claim 1 wherein the thrombospondin-1 is an isolated protein selected from the group consisting of: a protein isolated from human platelets, a fragment prepared from a protein isolated from human platelets, a recombinant human protein, a synthetic peptide, a recombinant human variant protein and a chimeric human protein.
5. (Original) The isolated complex according to Claim 1 wherein the heparin platelet factor 4 and thrombospondin-1 are present at a ratio determined to be optimal for recognition by

platelet factor 4/heparin/thrombospondin-1 ternary complex-reactive immunoglobulin present in a standardized positive control sample.

6. (Original) The isolated complex according to Claim 4 wherein the complex is preformed by combining 0.01-40  $\mu\text{g/ml}$  platelet factor 4, 0.01-1.0 U/ml unfractionated heparin and 0.01-40  $\mu\text{g/ml}$  thrombospondin-1.
7. (Original) The isolated complex according to Claim 5 wherein the complex is formed by combining 20  $\mu\text{g/ml}$  human platelet factor 4, 0.03 U/ml unfractionated heparin and 1  $\mu\text{g/ml}$  human thrombospondin.
8. (Withdrawn) A method for diagnosing type-2 heparin-induced thrombocytopenia comprising detecting the presence of platelet factor 4/heparin/thrombospondin-1 ternary complex-reactive immunoglobulin in a plasma or serum sample obtained from a patient receiving a heparin drug, wherein the presence of said immunoglobulin is indicative of type 2 heparin induced thrombocytopenia.
9. (Withdrawn) The method of Claim 8 wherein the heparin drug is selected from the group consisting of porcine intestinal mucosal heparin, bovine lung heparin, metal heparinates, heparinoids, low molecular weight heparin, and heparin-like compounds.
10. (Withdrawn) The method of Claim 9 wherein said ternary complex-reactive immunoglobulin is of a human isotype selected from the group consisting of IgM, IgA, IgG and any combination thereof.
11. (Withdrawn) A method of detecting the presence of platelet factor 4/heparin/thrombospondin-1 ternary complex reactive immunoglobulin in a biological sample, comprising the steps of :

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- (a) contacting the biological sample with an antigen complex comprising platelet factor 4/heparin/thrombospondin-1 ternary complex, thereby producing a first combination;
- (b) maintaining said first combination under conditions suitable to promote formation of antigen/antibody complexes referred to a first product;
- (c) contacting the product of step (b) with detectably-labeled anti-human immunoglobulin reactive reagent specific for at least one isotype of human immunoglobulin, thereby producing a second combination;
- (d) maintaining said second combination under conditions suitable to promote the binding of the detectably-labeled anti-human immunoglobulin reactive reagent to the antibody component of said first product; and
- (e) detecting the presence of the detectably-labeled anti-human immunoglobulin reactive reagent, wherein detection of the reagent demonstrates the presence of ternary complex reactive immunoglobulin in the biological sample.

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- 12. (Withdrawn) The method of Claim 11 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex comprises a heparin drug selected from the group consisting of porcine intestinal mucosal heparin, bovine lung heparin, a metal heparinate, a heparinoid, a low molecular weight heparin, a heparin-like compound and a combination of any of the preceding.
- 13. (Withdrawn) The method of Claim 11 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex comprises platelet factor 4 selected from the group consisting of a protein isolated from human platelets, a fragment prepared from a protein isolated from human platelets, a recombinant human protein, a synthetic peptide, a recombinant human variant protein, a chimeric human protein, and a combination of any of the preceding.
- 14. (Withdrawn) The method of Claim 11 wherein the thrombospondin-1 is an isolated protein selected from the group consisting of a protein isolated from human platelets, a

fragment prepared from a protein isolated from human platelets, a recombinant human protein, a synthetic peptide, a recombinant human variant protein and a chimeric human protein.

15. (Withdrawn) The method of Claim 11 wherein the antigen complex is preformed at a ratio determined to be optimal for recognition by platelet antigen complex-reactive immunoglobulin present in a standardized positive control sample.
16. (Withdrawn) The method of Claim 11 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex is immobilized on a solid support.
17. (Withdrawn) The method according to Claim 16 wherein the solid support is selected from the group consisting of: polycarbonate, polyallomer, polypropylene, polyvinyl, nylon, nitrocellulose, polystyrene and maleic anhydride activated polystyrene.
18. (Withdrawn) The method of Claim 11 wherein the ternary complex-reactive immunoglobulin has an isotype selected from the group consisting of: IgM, IgA, IgG and any combination thereof.
19. (Withdrawn) The method of Claim 11 wherein the detectably-labeled anti-human immunoglobulin reactive reagent specific for at least one isotype of human immunoglobulin comprises a reagent selected from the group consisting of: an anti-human immunoglobulin polyclonal antibody, an anti-human IgG polyclonal antibody, an anti-human IgM polyclonal antibody and an anti-human IgA polyclonal antibody.
20. (Withdrawn) The method of Claim 11 wherein the detectably-labeled anti-human immunoglobulin reactive reagent specific for at least one isotype of human immunoglobulin is selected from the group consisting of: intact immunoglobulin, F(ab)<sub>2</sub> fragments and F(ab) fragments.

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21. (Withdrawn) The method of Claim 11 wherein the detectably-labeled anti-human immunoglobulin reactive reagent comprises a detectable label selected from the group consisting of: an enzyme, a radioactive molecule, an affinity ligand and a fluorophore.
22. (Withdrawn) The method according to Claim 21 wherein the enzyme is selected from the group consisting of: horseradish peroxidase, alkaline phosphatase,  $\beta$ -galactosidase, glucose oxidase.
23. (Withdrawn) The method of Claim 21 wherein the affinity ligand is biotin.
24. (Withdrawn) A method for the identification of an individual at risk for the occurrence of a thrombotic complication of type 2 heparin-induced thrombocytopenia, comprising the steps of:
- (a) contacting a plasma or serum sample obtained from the individual with a platelet factor 4/heparin/thrombospondin-1 ternary complex, thereby producing a first combination;
  - (b) maintaining said first combination under conditions suitable to promote the formation of an antibody/antigen complex, wherein the antibody is derived from the plasma sample and the platelet factor 4/heparin/thrombospondin-1 ternary complex is the antigen; and
  - (c) detecting the antibody/antigen complex of step (b), wherein formation of the antibody/antigen complex is indicative of the individual being at risk for the occurrence of a thrombotic complication of type-2 heparin-induced thrombocytopenia.
25. (Withdrawn) The method of Claim 24, wherein detecting the presence of the antibody/antigen complex of step (b) is done by contacting the complex of (b) with a detectably labeled immunologic reagent specific for binding to human IgG, IgA, IgM or any combination thereof.

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26. (Withdrawn) The method of Claim 24 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex is immobilized on a solid phase.
27. (Withdrawn) The method according to Claim 24 wherein the solid phase is selected from the group consisting of: polycarbonate, polyallomer, polypropylene, polyvinyl, nylon, nitrocellulose, polystyrene and maleic anhydride activated polystyrene.
28. (Withdrawn) The method according to Claim 25 wherein said detectably labeled immunologic reagent is selected from the group consisting of: intact immunoglobulin, F(ab)<sub>2</sub> fragments and F(ab) fragments.
29. (Withdrawn) The method of Claim 24 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex comprises heparin selected from the group consisting of: porcine intestinal mucosal heparin, bovine lung heparin, a metal heparinate, a heparinoid, low molecular weight heparin, a heparin-like compound and a combination of any of the preceding.
30. (Withdrawn) The method of Claim 24 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex comprises platelet factor 4 selected from the group consisting of: a protein isolated from human platelets, a fragment prepared from a protein isolated from human platelets, a recombinant human protein, a synthetic peptide, a recombinant human variant protein, a chimeric human protein and a combination of any of the preceding .
31. (Withdrawn) The method of Claim 24 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex comprises thrombospondin-1 selected from the group consisting of: a protein isolated from human platelets, a fragment prepared from a protein isolated from human platelets, a recombinant human protein, a synthetic peptide, a recombinant human variant protein, a chimeric human protein and a combination of any of the preceding.

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32. (Withdrawn) The method of Claim 24 wherein the platelet factor 4/heparin/thrombospondin-1 ternary complex is preformed at a ratio determined to be optimal for the formation of a complex which is recognized by ternary complex-reactive immunoglobulin present in a standardized positive control sample.
33. (Withdrawn) The method of Claim 25 wherein the detectably labeled immunologic reagent comprises a label selected from the group consisting of: an enzyme, a radioactive molecule, an affinity ligand and a fluorophore.
34. (Original) A kit for an enzyme linked immunoadsorbent assay for detecting the presence of immunoglobulin reactive with a platelet factor 4/heparin/thrombospondin-1 antigen complex, comprising:
- a buffered medium comprising heparin;
  - a buffered medium comprising isolated human PF4;
  - a buffered medium comprising isolated human TSP-1;
  - a wash medium formulated to reduce nonspecific binding;
  - at least one anti-human immunoglobulin reactive reagent detectably labeled with a reporter molecule, and having a specificity for at least one isotype of human immunoglobulin;
  - a standardized positive control comprising known amounts of ternary complex reactive antibody;
  - a negative control sample;
  - a substrate for the reporter molecule; and
  - a diluent reagent.
35. (Original) The kit according to Claim 34 further comprising a solid phase support suitable for the immobilization of a platelet factor 4/heparin/thrombospondin-1 ternary complex, wherein said solid phase support comprises a material selected from the group consisting of: polycarbonate, polyallomer, polypropylene, polyvinyl, nylon, nitrocellulose, polystyrene and maleic anhydride activated polystyrene.

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36. (Original) A kit for detecting the presence of immunoglobulin reactive with a platelet factor 4/heparin/thrombospondin-1 antigen complex, comprising:
- a solid phase support material on which a platelet factor 4/heparin/thrombospondin-1 ternary complex has been immobilized; a buffered medium comprising isolated human TSP-1;
  - a wash medium formulated to reduce nonspecific binding;
  - at least one anti-human immunoglobulin reactive reagent detectably labeled with a reporter molecule, and having a specificity for at least one isotype of human immunoglobulin;
  - a standardized positive control comprising known amounts of ternary complex reactive antibody;
  - a negative control sample; and
  - a diluent reagent.
37. (Previously Presented) A kit for detecting the presence of immunoglobulin reactive with a platelet factor 4/heparin/thrombospondin-1 antigen complex, comprising:
- a solid phase support material on which a platelet factor 4/heparin/thrombospondin-1 ternary complex has been immobilized;
  - a wash medium formulated to reduce nonspecific binding;
  - at least one anti-human immunoglobulin reactive reagent detectably labeled with a reporter molecule, and having a specificity for at least one isotype of human immunoglobulin;
  - a standardized positive control comprising known amounts of ternary complex reactive antibody;
  - a negative control sample; and
  - a diluent reagent.
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